water, some of them fixed in 1 : 3 acetic ethanol for immediate testing. The rest was put to recovery and fixed at regular intervals upto 96 hr and root-tips were squashed in Feulgen's stain after hydrolysis. Treatment and recovery were carried out at 20 + 2°C along with controls.

The rate of mitosis was not affected appreciably by caffeine. It suppressed cell plate formation resulting in binucleate cells and induced clastogenic effects leading to chromosomal abnormalities. At concentrations higher than 50 mM caffeine, polyploid cells (figure 2) often with fragmented chromosomes and bridges were of predominant occurrence. A significant observation from the same samples was a special type of anaphasic grouping "meiotic reduction I" where homologous chromosomes segregated during mitotic anaphase. Though these groupings were not very frequent they presented a contrast to the high percentage of polyploid cells and could be spotted at once. Only those anaphases (figure 3) showing both qualitative and quantitative haploid complement were identified as "meiotic reductions". The highest percentage of such groupings scored was 13.04 in 50 mM/2 hr T/48 hrR. The frequency of "meiotic reductions" did not bear any relationship to the concentration of caffeine or the amount of recovery allowed after treatment. If this "meiotic reduction" simulates meiosis I, other stages comparable to meiosis II, identified from the same caffeine treated root-tips were "meiotic reduction II" (figure 4) and a "tetrad" (figure 5). Haploid cells (figure 6) probably resultant of cytokinesis after "meiotic reduction I" were observed in some instances. The appearance of all these three stages from samples with 24 hr or more of recovery time suggests their origin after 2 or more cell cycles. Occurrence of reductional groupings and haploid cells was reported by Huskins[6] with sodium nucleate treatment in *Allium* root-tips who referred to it as 'somatic meiosis' without however implying that all other features of meiosis also occur. The induction of "meiotic reductions" with treatments pyrimidine nucleotides, their intermediates and RNA in the same test system and also in *Pterotheca falconeri* was reported[7]. Caffeine, a purine derivative also is capable of inducing a similar effect lends support to the theory that it is the cumulative effect of purine and pyrimidine metabolism.

The author wishes to thank Prof. P. N. Mehra for valuable guidance.

8 July 1985; Revised 12 December 1985.

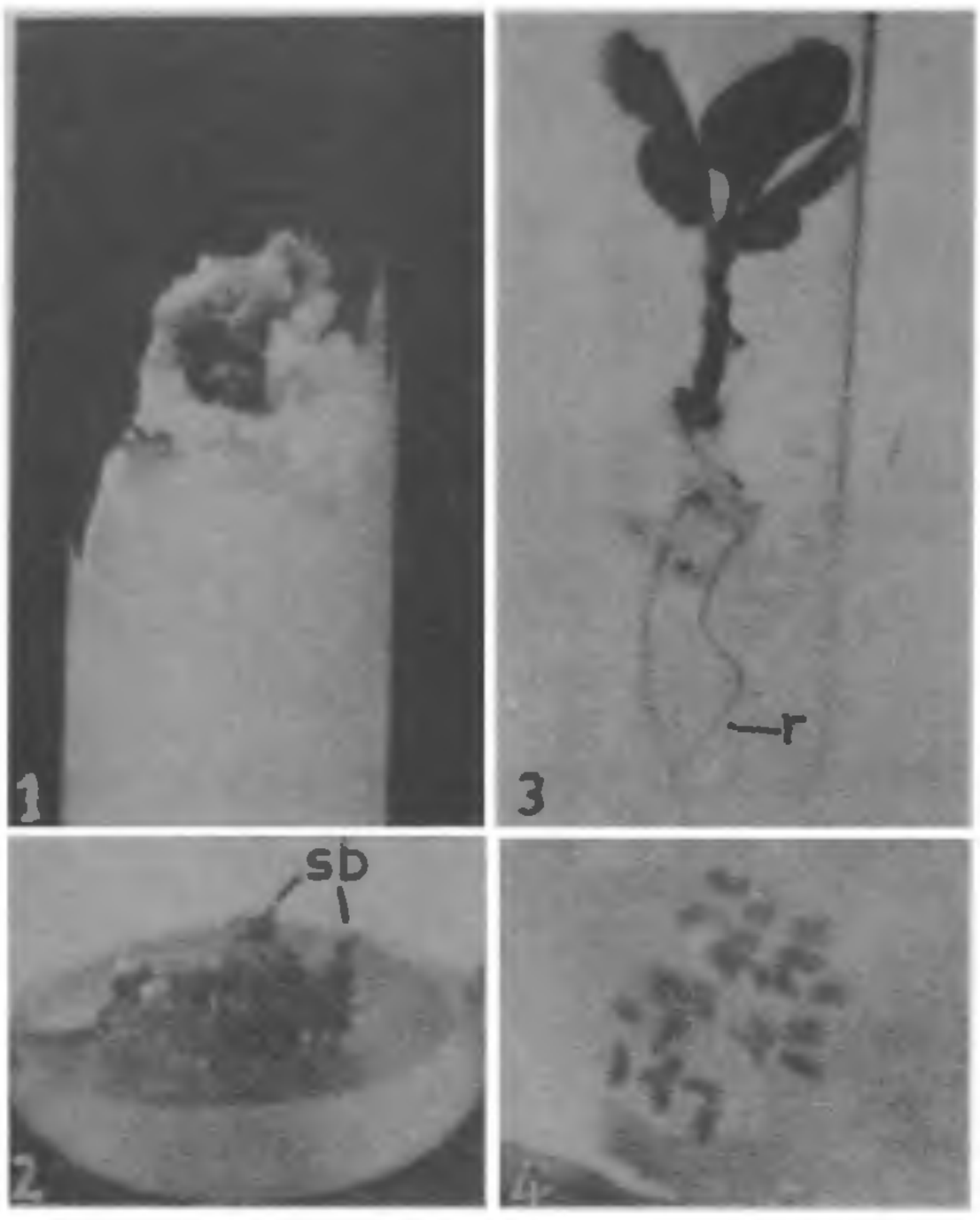
1. Kihlman, B. A., *Actions of chemicals on dividing cells*. Prentice Hall, New Jersey, 1966, p. 266.
2. Kihlman, B. A., *Mut. Res.*, 1974, **26,** 53
3. Kihlman, B. A. and Levan, A., *Hereditas*, 1949, **35,** 109.
4. Kihlman, B. A., Norlen, K., Sturlid, S. and Odmark, G., *Mut. Res.*, 1971, **12,** 463.
5. Ostertag, W., Dursberg, E. and Sturman, M., *Mut. Res.*, 1965, **2,** 293.
6. Huskins, C. L., *J. Hered.*, 1948, **39,** 311.
7. Kumari, L. and Mehra, P. N., *Nucleus*, 1981, **25,** 136.

REGENERATION OF PLANTS FROM LEAF CALLUS CULTURES OF *SOLANUM TORVUM* SWARTZ

PRATAP NARAYAN and V. S. JAISWAL
Department of Botany, Banaras Hindu University, Varanasi 221 005, India.

IN recent years the technique of tissue and organ culture has been effectively used in the mass multiplication of a number of plants of economic importance[1]. However, in a few cases the leaf material has been used as the source of tissue. *Solanum torvum* contains solasonine alkaloid and is currently considered to be a promising species as commercial source of this alkaloid. Vegetative propagation of *S. torvum* has not been possible by cuttings and graftings, limiting the clonal multiplication of high yielding strains. The present investigation was, therefore, undertaken to develop an efficient tissue culture method for plantlet formation from leaf tissues of young plants of *Solanum torvum* Swartz.

Leaves were taken from young plants growing under field conditions in the campus of Banaras Hindu University. They were washed in running tap water for about 30 min and then treated with 1 % solution (v/v) of Cetavlon (a detergent and antiseptic) for 5 min. After rinsing with distilled water several times, the leaves were surface-sterilized with 0.1 % (w/v) mercuric chloride solution for 5 min followed by a thorough washing with double-distilled water. Discs were punched from the leaf lamina with sterile cork borer (5 mm diameter) and single disc was inoculated in each culture vial containing solidified Murashige and Skoog's medium[2] (MS), supplemented with various concentrations and combinations of kinetin, IAA and 2,4-D. Cultures were maintained at 25 ± 2°C with 12 hr illumination at about 4000 lx. For each



Figures 1–4. 1. Proliferation of callus from cut margin of a leaf disc on MS + 2,4-D (1 mg/l) + kinetin (0.5 mg/l). Five-week old culture × 1.4. **2.** Induction of multiple shoot buds (sb) from leaf callus subcultured on MS + kinetin (1 mg/l). Four-week-old culture × 1.5. **3.** Rooted shoot on MS liquid medium with filter paper support. Eight-week-old culture. *r* = root × 0.6. **4.** Metaphage chromosomes ($2n = 24$) from a squash preparation of root tip of an *in vitro* formed plant × 1250.

treatment 24 cultures were raised and all experiments were repeated at least thrice.

The leaf discs cultured on MS containing 1.0 mg/l 2,4-D and 0.5 mg/l kinetin, produced callus from cut margin of the leaf disc after 4–5 weeks of culture (figure 1). Five-week old calli obtained from these cultures if subcultured on medium supplemented with 0.5, 1.0, 2.0 or 3.0 mg/l kinetin, grew well and formed a number of small green protuberances on their surfaces. After 4–5 weeks of subculture 8–12 shoot buds differentiated from the green tissues and developed into shoots (figure 2). The percentage of cultures showing shoot multiplication at 0.5, 1, 2 and 3 mg/l kinetin was 20, 27, 23 and 15 respectively. Application of 0.5, 1, 2 and 3 mg/l IAA alone stimulated only root formation from the subcultured calli. In combination with kinetin (0.5 to 2.0 mg/l) IAA at lower concentration suppressed shoot bud differentiation, and on medium containing 2 mg/l IAA and 3 mg/l kinetin the calli remained completely unorganized.

For rooting the *in vitro* formed shoots were isolated and transferred to a filter paper bridge on MS basal liquid medium. Roots emerged directly from the base of shoot after 2 weeks of transfer resulting in the establishment of complete plantlet (figure 3). Diploid nature ($2n = 24$) of *in vitro* formed plants was established by examination of acetocarmine squashes of root tips (figure 4).

The present study clearly demonstrates the possibility of vegetative multiplication of *S. torvum* in cultures via shoot differentiation from leaf callus.

25 October 1985; Revised 11 December 1985

1. Flick, C. E., Evans, D. A. and Sharp, W. R., In: *Handbook of plant cell culture*, (eds) D. A. Evans, W. R. Sharp, P. V. Ammirato and Y. Yamada, Collier Macmillan Publishers, London, 1983, Vol. 1, p. 13.
2. Murashige, T. and Skoog, F., *Physiol. Plant.*, 1962, **15**, 437.

BIOCHEMICAL ACTIVITIES OF SOME THERMOPHILIC FUNGI

B. R. RENUKA, A. SUBRAHMANYAM*, M. VINAYAKUMAR**, K. PHANI KUMAR** and P. RAMA RAO
*Department of Botany, **Department of Microbiology, Osmania University, Hyderabad 500 007, India.*
**Department of Botany, Kakatiya University, Warangal 506 009, India.*

IN recent years, there has been considerable interest in utilizing thermophilic fungi in different industrial fermentation processes because of several advantages that high temperature fermentation affords. There are many reports on enzyme production[1–5] and antimicrobial activities[6, 7], but none on organic acids and phenolic compounds produced by thermophilic fungi. An attempt was therefore made to study the extracellular products of four thermophilic fungi *viz*, *Nodulisporium thermoroseum* A Subrahm and